Amendments to the Claims

This listing of claims will replace all prior versions, and listings, of claims in the application.

Listing of Claims:

1. (Currently Amended) A method for diagosing breast cancer in a subject, comprising:
   a) providing a plasma sample from ~~a~~ the subject, said plasma sample comprising genomic DNA, wherein said genomic DNA comprises a plurality of promoters from different genes;
   b) isolating and digesting said genomic DNA ~~in said plasma sample~~ with a methylation sensitive restriction enzyme under conditions such that unmethylated CpG islands in said promoters are cleaved while methylated CpG islands in said promoters are not cleaved;
   c) contacting said ~~plasma sample~~ digested genomic DNA with at least five different pairs of gene specific primers, wherein said gene specific primers are configured to hybridize to said genomic DNA and amplify five different promoters from at least five different genes including DAPK, and wherein said contacting is under conditions such that fragments of said plurality of promoters comprising uncleaved CpG islands are amplified, while cleaved promoters comprising cleaved CpG islands are not amplified; and
   d) detecting the presence or absence of DNA methylation in each of said plurality of promoters based on the amplification, or lack of amplification, of said fragments to generate a methylation profile for said subject, thereby diagnosing breast cancer in the subject.

2. (Currently Amended) The method of ~~Claim~~ claim 1, wherein said method further comprises comparing said methylation profile to ~~more~~ one or more standard methylation profiles, wherein said standard methylation profiles are selected from the group consisting of methylation profiles of non-cancerous samples and methylation profiles of cancerous samples.

3. (Currently Amended) A method of characterizing breast cancer in a subject, comprising:

a) providing a ~~biological~~ plasma sample from ~~a~~ the subject ~~diagnosed with breast cancer~~, said biological sample comprising genomic DNA; and

b) detecting the presence or absence of DNA methylation in a plurality of different genes including DAPK ~~and Progesterone Receptor~~, thereby characterizing breast cancer in the subject.

4. (Currently Amended) The method of claim 3, ~~further comprising the step of detecting the presence or absence of DNA methylation in at least one additional gene selected from the group consisting of S100A2, SRBC, BRCA1, HIN1, Cyclin D2, TMS1, HIC-1, hMLH1, E-cadherin, 14-3-3sigma, GSTP, p15, MDR1, Calcitonin, RIZ1, RARbeta, and MDGI~~ wherein the plurality of different genes include a gene selected from the group consisting of FAS, MCT1, p16, PAX5, THBS, TRANCE, and VHL.

5. (Currently Amended) The method of claim 3, wherein said characterizing breast cancer comprises ~~detecting the presence or absence of chemotherapy resistant cancer~~ diagnosing breast cancer.

6.-11. (Cancelled)

12. (Original) The method of claim 3, wherein said DNA methylation comprises CpG methylation.

13. (Currently Amended) The method of claim 3, wherein said detecting the presence or absence of DNA methylation comprises the digestion of said genomic DNA with a methylation-sensitive restriction enzyme followed by multiplexed amplification of gene-specific DNA fragments ~~with~~ having CpG islands.

14. (Currently Amended) The method of claim 13, wherein said methylation-sensitive restriction enzyme comprises ~~Hin61~~ Hin6I.

15.-22. (Canceled)

23. (Previously Presented) The method of claim 1, wherein said DNA methylation comprises CpG methylation.

24. (Currently Amended) The method of claim 1, ~~wherein said cancer is breast cancer~~ wherein said gene specific primers are configured to hybridize to said genomic DNA and amplify five different promoters from at least five different genes including DAPK and a gene selected from a group consisting of FAS, MCT1, p16, PAX5, THBS, TRANCE, and VHL.

25. (Currently Amended) The method of Claim 1, wherein said methylation-sensitive restriction enzyme comprises ~~Hin61~~ Hin6I.

26. (Previously Presented) The method of Claim 1, further comprising the step of i) separating said plasma sample into a control sample and an experimental sample, and ii) adding control nucleic acid to both said control and experimental samples, wherein said control nucleic acid comprises at least one known CpG island that is unmethylated.

27. (Previously Presented) The method of Claim 26, wherein said control sample is not exposed to said digesting and said experimental sample is exposed to said digesting, and wherein both said control and experimental samples are contacted with primers specific for said control nucleic acid under conditions such that a fragment of said control nucleic acid is amplified only if said known CpG island is uncleaved.

28. (Previously Presented) The method of Claim 27, further comprising comparing any fragments amplified in said control and experimental samples to confirm that said digesting in step b) is complete.

29.-30. (Cancelled)

31. (Previously Presented) The method of Claim 1, wherein said digesting is performed to completion.

32. (Cancelled)

33. (Currently Amended) A method for diagosing breast cancer in a subject, comprising:

a) providing a biological sample from ~~a~~ the subject, said biological sample comprising genomic DNA, wherein said genomic DNA comprises a plurality of promoters from different genes;

b) adding control nucleic acid to said biological sample and separating said biological sample into a control sample and an experimental sample, wherein said control nucleic acid comprises at least one known CpG island that is unmethylated;

c) isolating said genomic DNA from said experimental sample and said control sample, and digesting said genomic DNA from said experimental sample, but not said genomic DNA from said control sample, with a methylation-sensitive restriction enzyme under conditions such that: i) unmethylated CpG islands in said promoters are cleaved while methylated CpG islands in said promoters are not cleaved, and ii) unmethylated CpG islands in said control nucleic acid are cleaved;

d) contacting said genomic DNA from said control and experimental samples with primers specific for said control nucleic acid and detecting the presence or absence of amplification of said genomic DNA from ~~in~~ said experimental and control samples, wherein no detectable amplification of said genomic DNA from ~~in~~ said experimental sample confirms digestion was complete, and wherein amplification of said genomic DNA from ~~in~~ said control sample confirms proper amplification of uncleaved control nucleic acid;

e) contacting said genomic DNA from said experimental sample with gene specific primers for a plurality of different genes including DAPK, wherein said gene specific primers are configured to hybridize to said genomic DNA, and wherein said contacting is under conditions such that fragments of said

plurality of promoters comprising uncleaved CpG islands are amplified, while cleaved promoters comprising cleaved CpG islands are not amplified; and

f) detecting the presence or absence of DNA methylation in each of said plurality of promoters of said genomic DNA of ~~is~~ said experimental sample based on the amplification, or lack of amplification, of said fragments to generate a methylation profile for said subject, thereby diagnosing breast cancer in the subject.

34. (New) The method of claim 34, wherein said gene specific primers are configured to hybridize to said genomic DNA and amplify different genes including DAPK and a gene selected from a group consisting of FAS, MCT1, p16, PAX5, THBS, TRANCE, and VHL.